62. A method for inhibiting the proliferation of a malignant cell line that expresses the PBR gene, comprising introducing into said cell line an antisense oligonucleotide according to claim 57 in an amount effective to inhibit cell proliferation.

63. The antisense oligonucleotide of claim 53, which is comprised in a proteoliposome containing viral envelope receptor proteins.

64. The antisense oligonucleotide of claim 53, which comprises part of a vector.

65. The antisense oligonucleotide of claim 53, which is comprised in a vector which is expressed in the mammary gland.

66. The antisense oligonucleotide of claim 53, which is contained in a carrier.

67. The antisense oligonucleotide of claim 66 wherein said carrier is a protein selected from the group consisting of a cytokine or polylysine-glycoprotein carrier.

68. The antisense oligonucleotide of claim 53, which is comprised in a microbead.--

REMARKS

Entry of the foregoing amendments, reconsideration reexamination of the subject application, as amended, pursuant to and consistent with 37 C.F.R. § 1.112, and in light of the remarks which follow, are respectfully requested. By the present amendments, claims 48 to 52 have been cancelled without prejudice in favor of new claims 53-68. These amendments are introduced in an effort to expedite prosecution.

Prior to specifically addressing the rejections, the Examiner is respectfully thanked for the interview held with the undersigned, Samir Elamrani and the Examiner on October 11, 2000. During that interview, all of the outstanding issues were discussed, however the 112 enablement and written description issues were discussed in particular detail. Essentially, applicants' representatives strenuously argued that the specification provides sufficient information to enable and describe to one of ordinary skill how to construct antisense oligonucleotides that would inhibit the expression of a PBR gene when introduced into a cell line that expresses said gene, and would consequently inhibit cell proliferation. In support thereof, Applicants' representative referred to the data contained in the disclosure which indicates that by blocking the expression of one allele of the PBR gene it resulted in reduced cell proliferation of a cancerous cell line, particularly a human breast cancer cell line. Moreover, applicants' representative argued with respect to the written description issue, that the specification provides partial cDNA sequences for the human PBR gene, which would provide sufficient description to enable one and to describe to one of ordinary skill how to construct sequences having a complementary antisense structure, and furthermore that it would not require undue experimentation to select those of which sequences are effective, namely those which inhibit PBR expression and thereby inhibit cell proliferation.

Moreover, Applicants' representative further expressed the intent to submit additional method claims directed to using the subject antisense oligonucleotide in order to inhibit proliferation of particular cell lines. It was indicated that entry of these claims should not raise any significant new issues, particularly since the Examiner has already cited references which relate to the use of antisense and oligonucleotides in order to inhibit gene expression.

Also, applicants' representative indicated that the antisense oligonucleotides and the method of using such oligonucleotides to inhibit gene expression constitute a single unitary invention, and that these claims should properly be examined together.

Turning now to the Office Action, applicants note that claims 48 to 52 stand rejected under 35 U.S.C. § 112, first paragraph. This rejection is respectfully traversed to the extent it may be applicable to the claims as amended. At the outset, it is respectfully submitted that this rejection is wholly inconsistent with the anticipatory and § 103 obviousness rejection made by the Examiner. Notwithstanding the fact that she alleges that the claims would have been obvious to one of ordinary skill in the art, she also alleges that there is insufficient information in the disclosure to enable the claimed invention. Applicants respectfully submit that these rejections cannot be reconciled. Moreover, for the reasons set forth below, neither the prior art rejection or the enablement rejections should be maintained against the current claims.

Specifically, the issue with respect to the alleged unpredictability associated with homologous recombination is respectfully traversed. While applicants acknowledge that it can not be reasonably predicted apriori what particular antisense oligonucleotides will be effective, namely inhibit PBR gene expression, it is predictable that sequences can be selected absent under experimentation which will be effective. The predictability of the outcome is established based on the information in the disclosure which demonstrates that an inhibition of the expression of one allele of the PBR gene resulted in a reduction in cell proliferation of a cancerous cell line. Therefore, the correlation between an inhibition of PBR expression and cell proliferation of tumor cells is contained in the disclosure. Specifically,

this information is contained in Example 7 which bridges pages 45-46 of the subject application. As indicated above, this example provides evidence which demonstrates that inactivation of one allele of the PBR gene resulted in the suppression of PBR mRNA binding and expression, and that this inhibition resulted in a decrease in the rate of cell proliferation (determined using the MTT proliferation assay which is an accepted method for measuring cell proliferation). Also, the example demonstrates that this reduced cell proliferation was in comparison to an appropriate control, namely a wild type cell, which does not contain a disruption of such gene. Therefore, the specification clearly contains evidence which establishes that inhibiting the expression of PBR will predictably result in reduced cell proliferation. While applicant agrees that it cannot be predicted at the outset what particular antisense oligonucleotides will be effective, it is not unpredictable that some can be selected that will work.

Moreover, as discussed at the interview, applicants are in the process of obtaining further information in order to further bolster the enablement of the claims. While applicants respectfully believe that there is sufficient information to enable the claims, supplemental information may be subsequently provided in the form of § 132 Declaration by the inventors. This information will be provided as soon as it becomes available.

Also, applicants respectfully submit that the 112 enablement rejection with respect to the unpredictability of antisense therapy, should not be maintained as this is not required to enable or describe the claim. While applicants agree that this is certainly a significant commercial and practical application of the claimed oligonucleotides, it is not the only utility of the claimed sequences. Indeed, as described in the subject application, the subject

antisense oligonucleotides may be utilized in order to identify cell lines wherein the proliferation thereof correlates to the expression of the PBR gene. This will provide a significant diagnostic utility, as the expression of PBR and its effect on cell proliferation correlates to the invasive and chemotactic potential of certain cell lines, particularly breast tumor cell lines. In fact, as discussed in the subject application, a partial sequence analysis has revealed that a particular PBR sequence which is contained in SEQ ID NO: 1 and 2, which is comprised in several breast cancer cell lines, i.e., MDA-231 and MCF-cells, of which MDA-231 is a highly aggressive breast cancer cell lines, suggest that the expression of these sequences may represent an early event in the progression of the disease. Also, data in the subject application indicate that nuclear PBR is responsible for regulating the movement of cholesterol into the nuclear membrane and that this regulation is related to the modulation of cell proliferation. Therefore, based on the foregoing withdrawal of the outstanding 112 enablement rejection is respectfully requested.

Also, previous claim 48 was rejected under 35 U.S.C. § 112, first paragraph as being broader than the enabling disclosure. Obviously, this should be moot as the claims are no longer directed to any non-naturally occurring compound which inhibits or reduces the expression of PBR.

Claims 48-52 further were rejected under 35 U.S.C. § 112, first paragraph on written description basis. This rejection is respectfully traversed to the extent it may be applicable to the claims as amended. As discussed above, and reflected by the present claim amendments, applicants have rewritten the claims to recite that the claimed antisense oligonucleotide possesses a complementary structure to at least a portion of the PBR cDNA contained in SEQ

ID NO: 1 or ID NO: 2. As may be seen upon review of the sequences, they are relatively short nucleic acid sequences. Particularly SEQ ID NO: 1 contains 652 base pairs as does SEQ ID NO: 2. Therefore, it would be readily apparent to one of skill in the art upon review of the subject application, who is further aware of antisense DNA techniques, what particular nucleic acid sequences would potentially fall within the scope of the claimed invention. Essentially, the invention would potentially embrace any sequence which possesses a complementary antisense structure to a portion of either of the sequences, which further must possess a particular functional characteristic, namely when introduced into a cell line that expresses PBR, the sequence must be able to inhibit the expression of said gene, and thereby inhibit cell proliferation. While applicants agree that it cannot be absolutely predicted what particular complementary sequences will be effective, applicants do not agree that it cannot be envisioned what is the genus of sequence based on the information in the disclosure. Indeed, with SEQ ID NO: 1 or 2 in hand, one skilled in the art would readily be able to know what population of nucleic acid sequences would potentially fall within the scope of the claims, and moreover it would be routine to select from those sequences those which inhibit gene expression. In this regard, the specification contains convincing data which demonstrates that inhibition of PBR expression results in reduced cell proliferation. Therefore, applicants respectfully submit that is clear from the disclosure that applicants were in possession of the invention as of the filing date, for significant purposes of satisfying the written description inquiry. Contrary to what the Examiner states in the Office Action, the specification does clearly allow persons of ordinary skill in the art to recognize what is claimed. In fact, it is apparent that the Examiner also is aware of what is claimed based on

her citation of 102 and 103 references. Indeed, the written description rejection is also inconsistent with the anticipatory and obviousness rejection. Clearly, there is no ambiguity as to what is being claimed, especially given the high level of the state of the art. Therefore, based on the foregoing, withdrawal of the §112 written description rejection is respectfully requested.

Claims 48, 49, 51 and 52 stand rejected under 35 U.S.C. § 102(a) as assertedly being anticipated by Papadopoulous et al. This reference relates to a replacement vector which provides for homologous recombination which disrupts the expression of PBR gene by means of recombination with the PBR gene and the genome. Based on the previous claims, the Examiner indicated that this reference anticipated the claimed compounds. However, it is respectfully submitted that this rejection should be moot in light of the present amendments. In particular, the claims are now directed to an antisense DNA which disrupts expression of PBR by a different mechanism than the reference. Consequently, the § 102 rejection should no longer be applicable to the current claims. Indeed, previous claim 50 which corresponded to an antisense nucleic acid sequence was not included in the previous anticipatory rejection. Therefore, the Examiner has previously at least implicitly concluded that the reference would not anticipate the claimed invention. Withdrawal of this rejection is therefore respectfully requested.

Claim 48 stands rejected under 35 U.S.C. § 102(a) as assertedly being anticipated by Moser et al. or Garnier et al. This rejection is also moot, based on the present amendments. In particular, the rejection while was applied based on its disclosure relating to a compound which inhibited the expression of PBR. However, as neither of these references teaches or

suggests an antisense nucleic acid sequence which inhibits the expression of PBR, this rejection is not applicable against the current claims. Withdrawal of the rejection therefore is respectfully requested.

Claims 48-51 further stand rejected under 35 U.S.C. § 103(a) as being unpatentable over Casalotti Gene (1992) in view of Weiss U.S. Patent 5,840,708 and Lu C et al. *Clin. Cancer Res.* 2(8): 1417-1425 (1996). Essentially, the position of the Examiner is that the primary reference Casalotti teaches the structure of rat PBR and Weiss teaches how to make antisense oligonucleotides, and the use thereof is therapeutics in order to inhibit the expression of a particular gene, and that Lu et al. teaches clones which are transfected with antisense DNA expression vector, in order to inhibit the expression of this gene. Based thereon, the Examiner alleges that it would have been obvious to have made antisense oligonucleotides to any known nucleic acid sequence. This rejection is respectfully traversed.

At the outset, it is noted that the claims have been refocused such that they are now directed to antisense oligonucleotides which inhibit the expression of the PBR gene, wherein the antisense nucleic acid sequence is complementary to the nucleic acid sequence contained in SEQ ID NO: 1 or 2. This sequence corresponds to a portion of human PBR. Therefore, even assuming for the sake of argument that the rejections were proper, the cited references would not teach or suggest the claimed invention.

Moreover, applicants respectfully traverse the rejection even as it pertains to the previous claims. Essentially, the rejection is improper because absent the recognition of the particular effect of PBR, namely that this portion has a regulatory effect on cell proliferation, particularly the proliferation of some tumor cell types, it would not have been obvious to

have constructed an antisense nucleic acid sequence in order to inhibit the expression of such gene. Only upon the demonstration of the modulatory effect of PBR on cell proliferation and malignant phenotype, would it have been obvious to have constructed such antisense oligonucleotides. Therefore, for both of these reasons, namely that the references does not teach or suggest an antisense DNA to the particular PBR sequence which is the subject of the present invention and further does not suggest the effect of PBR on tumor phenotype, the rejection should be withdrawn.

Based on the foregoing, this application is believed to be in condition for allowance. A notice to that effect is respectfully solicited. However, if any issues remain outstanding, the Examiner is respectfully requested to contact the undersigned so that prosecution of this application may be expedited.

Respectfully submitted,

SHAW PITTMAN

By: _____
Robin L. Teskin
Registration No. 35,030

2300 N Street, N.W.
Washington, D.C. 20037-1128
(202) 663-8000

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